**S510 (k) SUBSTANTIAL EQUIVALENCE DETERMINATION**
**DECISION SUMMARY**
**ASSAY AND INSTRUMENT COMBINATION TEMPLATE**

**A. 510(k) Number:**

K053571

**B. Purpose for Submission:**

New Device

**C. Measurand:**

Hemoglobin A, A2, F, S, C, E, D

**D. Type of Test:**

Quantitative, Semi-quantitative, Qualitative

**E. Applicant:**

Interlab Scientific Instruments srl

**F. Proprietary and Established Names:**

Microgel Alkaline Hemoglobin Electrophoresis test system
Microgel Acid Hemoglobin Electrophoresis test system

**G. Regulatory Information:**

1. Regulation section:

   21 CFR 864.7440, Electrophoretic Hemoglobin Analysis System
   21 CFR 864.7415, Abnormal Hemoglobin Assay

2. Classification:

   Class II

3. Product code:

   JBD, System, Analysis, Electrophoretic Hemoglobin

GKA, Abnormal Hemoglobin Quantitation

4. Panel:

   Hematology (81)

## H. Intended Use:

1. Intended use(s):

   The Microgel Alkaline Hemoglobin Electrophoresis test kit is intended for qualitative and semi-quantitative determination of normal hemoglobins (A1, A2 and F) as well as certain abnormal or variant hemoglobins (S or D and C or E) using agarose gel. To distinguish hemoglobins S from D or C from E an alternate confirmatory test such as acid hemoglobin electrophoresis is necessary. The electrophoretic test is performed at alkaline pH and provides a valuable screening method for hemoglobin patterns. Densitometry of the pattern allows the relative quantification of hemoglobin bands. The kit SRE604K has been designed for use with the fully automated Microgel instrument.

   The Microgel Acid Hemoglobin Electrophoresis kit is a qualitative test for the identification of both normal and abnormal or variant hemoglobins, and to confirm the identity of clinically relevant hemoglobins such as A, F, S and C. The Acid Hemoglobin test kit employs agarose gel at acidic pH and is for *in vitro* diagnostic use. The kit has been designed for use with the fully automated Microgel instrument.

2. Indication(s) for use:

   The Microgel Alkaline Hemoglobin Electrophoresis test kit is intended for qualitative and semi-quantitative determination of normal hemoglobins (A1, A2 and F) as well as certain abnormal or variant hemoglobins (S or D and C or E) using agarose gel. To distinguish hemoglobins S from D or C from E an alternate confirmatory test such as acid hemoglobin electrophoresis is necessary. The electrophoretic test is performed at alkaline pH and provides a valuable screening method for hemoglobin patterns. Densitometry of the pattern allows the relative quantification of hemoglobin bands. The kit SRE604K has been designed for use with the fully automated Microgel instrument.

   The Microgel Acid Hemoglobin Electrophoresis kit is a qualitative test for the identification of both normal and abnormal or variant hemoglobins, and to confirm the identity of clinically relevant hemoglobins such as A, F, S and C. The Acid Hemoglobin test kit employs agarose gel at acidic pH and is for *in vitro* diagnostic use. The kit has been designed for use with the fully automated Microgel instrument.

3. Special conditions for use statement(s):

   Not applicable.

4. Special instrument requirements:

   Not applicable.

## I. Device Description:

The Microgel Alkaline and Acid Hemoglobin Electrophoresis test kits consist of seven components; (1) Agarose gel plates(2) Buffered Sponges, (3) Lysing solution, (4) Amino Black Stain Solution, (5) Washing Solution for Applicators, (6) Disposable Sample Plates, (7) Blotters. The Microgel Alkaline and Acid Hemoglobin Electrophoresis test kits are used in conjunction with a Microgel instrument, a laboratory analyzer for electrophoretic analysis. The electrophoresis is performed using the migration phenomena on Mylar membranes, on which agarose gel is deposited (GEL), when voltage is applied. The Microgel Unit, with external command from a PC, sets up a sequence of basic phases to conduct the analysis. The instrument monitoring during the analysis is performed by the external PC with a bidirectional serial link.

1. Predicate device name(s):

   Sebia HYDROGEL HEMOGLOBINS
   Interlab Alkaline Hemoglobin
   Interlab Acid Hemoglobin

2. Predicate 510(k) number(s):

   K991362
   K032862
   K040146

3. Comparison with predicate:

| **Similarities** | | | |
|---|---|---|---|
| Item | ***MICROGEL HEMOGLOBINS*** | ***SEBIA HYDRAGEL HEMOGLOBINS*** | ***MICROTECH HEMOGLOBINS*** |
| **Intended use** | The Microgel Alkaline Hemoglobin Electrophoresis test kit is intended for qualitative and semi-quantitative determination of normal hemoglobins (A1, A2 and F) as well as certain abnormal or variant hemoglobins (S or D and C or E) using agarose gel. To distinguish hemoglobins S from D or | The Hydragel Hemoglobin is designed for separation of normal hemoglobins (A1 and A2) and for the detection of the major hemoglobin variant: S or D and C or E by electrophoresis on alkaline agarose gels. The resulting electrophoregrams are evaluated visually for | The Interlab Alkaline Hemoglobin Electrophoresis test system is intended for the separation of normal hemoglobins (A1, A2 and F) as well as certain abnormal or variant hemoglobins (S or D and C or E) using cellulose acetate supported on Mylar. The test is a screening method for in vitro diagnostic use in the Microtech 672 PCPC and the Microtech 648 |

| Similarities | | | |
|---|---|---|---|
| Item | ***MICROGEL HEMOGLOBINS*** | ***SEBIA HYDRAGEL HEMOGLOBINS*** | ***MICROTECH HEMOGLOBINS*** |
| | C from E an alternate confirmatory test such as acid hemoglobin electrophoresis is necessary. The electrophoretic test is performed at alkaline pH and provides a valuable screening method for hemoglobin patterns. Densitometry of the pattern allows the relative quantification of hemoglobin bands. The kit SRE604K has been designed for use with the fully automated Microgel instrument.<br><br>The Microgel Acid Hemoglobin Electrophoresis kit is a qualitative test for the identification of both normal and abnormal or variant hemoglobins, and to confirm the identity of clinically relevant hemoglobins such as A, F, S and C. The Acid Hemoglobin test kit employs agarose gel at acidic pH and is for *in vitro* diagnostic use. The kit has been designed for use with the fully automated Microgel instrument. | pattern abnormalities. Densitometry can serve as an aid in the interpretation by providing relative concentrations of individual fractions.<br><br>The Hydragel Hemoglobins are designed for separation of normal hemoglobin A, abnormal hemoglobins S and C and fetal hemoglobin F, by electrophoresis on acidic agarose gels. Hydragel Acid Hemoglobin is essential to confirm the identification of hemoglobin variants previously detected on alkaline gels, in particular to differentiate hemoglobins S from D and E from C. The tests are used in conjunction with the semi-automated Hydrasys system. | ISO fully automated analyzers. To distinguish hemoglobins S from D or C from E an alternate confirmatory test such as acid hemoglobin electrophoresis is necessary.<br><br>The Interlab Acid Hemoglobin Electrophoresis test system is intended for the electrophoresis separation of hemoglobins to confirm the identity of clinically relevant hemoglobins such as A, F, S, and C. It is to be used in conjunction with the Interlab Alkaline Hemoglobin Electrophoresis test kit. The Acid Hemoglobin test kit is for in vitro diagnostic use and can be automated on the Microtech 672 PCPC and Microtech 648 ISO instruments. |
| **Results** | Visual | Same | Same |
| **Method** | Agarose gel electrophoresis | Agarose gel electrophoresis | Cellulose Acetate or Mylar |
| **Test Principle** | Normal hemoglobins and most variant have different electrophoretic mobility and can be resolved according to their net charge by | Normal hemoglobins and most variant have different electrophoretic mobility and can be resolved according to their net charge by | Normal hemoglobins and most variant have different electrophoretic mobility and can be resolved according to their net charge by electrophoresis on cellulose |

| Similarities | | | |
|---|---|---|---|
| Item | ***MICROGEL HEMOGLOBINS*** | ***SEBIA HYDRAGEL HEMOGLOBINS*** | ***MICROTECH HEMOGLOBINS*** |
| | electrophoresis on agarose gel in alkaline or acid buffer. | electrophoresis on agarose gel in alkaline or acid buffer. | acetate in alkaline or acid buffer. |
| **Specimen** | Hemolyzed separated red blood cells using hemolyzing solution. | Hemolyzed separated red blood cells using hemolyzing solution | Hemolyzed separated red blood cells using distilled water |
| **Sample Application** | Pipetting Station | Same | Same |
| **Detection** | Densitometric | Same | Same |

| Differences | | | |
|---|---|---|---|
| Item | ***MICROGEL HEMOGLOBINS*** | ***SEBIA HYDRAGEL HEMOBLOBINS*** | ***MICROTECH HEMOGLOBINS*** |
| **Equipment and Accessories** | Microgel System<br><br>Alternative to Microgel systems:<br>Migration Chamber and power supply<br>Sample applicator<br>Reagent containers<br>Dryer<br>Optional Densitometer | Sebia Hydrasys System<br><br>Alternative to Hydrasys System:<br>Sebia Migration Cell<br>Sebia Sample Applicator<br>Sebia Gel Processing/Staining Module<br>Optional Densitometer | Microtech 648 ISO System<br>Microtech 672 PCPC System<br><br>Alternative to Microtech systems:<br>Migration Chamber and power supply<br>Sample applicator<br>Reagent containers<br>Dryer<br>Optional Densitometer |
| **Reagents** | Alkaline Hb Gel Plates<br>Acid Hb Gel Plates<br>Buffered Sponges<br>Lysing Solution<br>Amino Black Staining<br>Washing Solution for Applicators | Alkaline Hb Gel Strips<br>Acid Hb Gel Strips<br>Buffered Strips<br>Hemolysing Reagent<br>Staining Solution<br>Diluent<br>Amidoblack Stain | Alkaline Hb Gel Strips<br>Acid Hb Gel Strips<br>Running Buffer<br>Destaining Solution<br>Clearing Solution<br>Ponceau Red Stain |

## K. Standard/Guidance Document Referenced (if applicable):

## L. Test Principle:

*Microgel Alkaline Hemoglobin Electrophoresis*
Normal hemoglobins and most variants have different electrophoretic mobility and can be resolved according to their net charge by electrophoresis on agarose gel plate in an alkaline buffer. The analysis is performed using the fully automated Microgel instrument. When the electrophoretic separation of the bands is complete, the agarose gel plate is denatured, stained with Amino Black, distained and dried. The patters are then scanned and the densitometric results are shown together with the graph.

*Acid Hemoglobin Electrophoresis*
Normal hemoglobins and most variants have different electrophoretic mobility and can be resolved according to their net charge by electrophoresis on agarose gel in acid buffer. This method allows separation of those variant hemoglobins that overlap in the alkaline electrophoretic pattern. The analysis is performed using the fully automated Microgel instrument. When the electrophoretic separation of the bands is complete, the agarose gel plate is denatured, stained with Amido Black, destained and dried. Visual inspection of the patterns is performed to detect both normal and abnormal or variant hemoglobins.

## M. Performance Characteristics (if/when applicable):

1. Analytical performance:

*a. Precision/Reproducibility:*

Microgel Alkaline Hemoglobin – Intra-run
Three different samples were run on Microgel; thirteen replicates of each sample were run on one gel plate using the same batch number of reagents. Electrophoretic patterns were evaluated visually. Mean, SD, and CV values were calculated for each fraction.

**Precision (Intra-run) results:**

| Sample # | Hb A | Hb A2 | Hb F | Hb S | Hb C | Mean (%) | SD | CV (%) |
|---|---|---|---|---|---|---|---|---|
| A | X | | | | | 62.8 | 0.87 | 1.38 |
| | | | | | X | 37.2 | 0.87 | 2.33 |
| B | X | | | | | 57.12 | 0.12 | 0.2 |
| | | X | | | | 1.26 | 0.05 | 4.01 |
| | | | X | | | 31.15 | 0.19 | 0.62 |
| | | | | X | | 10.46 | 0.18 | 1.68 |
| C | X | | | | | 97.45 | 0.07 | 0.07 |
| | | X | | | | 2.55 | 0.07 | 2.59 |

*Microgel Acid Hemoglobin – Intra-run*
Three different abnormal samples (S, AFSA2, and AFSC) were used. Thirteen replicates of each sample were run on the sample gel plate, using the same batch number of the kit. The electrophoretic patterns were inspected visually. The presences of all abnormal bands were confirmed.

*b. Linearity/assay reportable range:*

An abnormal patient sample was serially diluted and run on the Interlab Hemoglobin test system assess the linearity of the system. Hemoglobin A1 and A2 bands were measured to show the range of concentrations, and assess the lowest concentration that can be detected on the agarose gel system. The results are as follows:

Linearity range for HbA2 in emolised sample: 0.4 – 2.1 g/L
(Total Hb concentration: 150 g/L)
% of HbA2 = 5.6%  Conc. HbA2 = 8.4 g/L

*c. Traceability, Stability, Expected values (controls, calibrators, or methods):*

*Microgel Alkaline Hemoglobin*
Stability studies were performed at two time periods Test O (time O), Test 1 (after 6 months from production date). During these periods the kits was stored at 15 – 30$^{o}$C. Used in this study were control solution AFSC and three different normal patients (two containing HbA1 and HbA2 and carbonic anhydrase, and one contain traces of HbF).

*Microgel Acid Hemoglobin*
Stability studies were performed at three time periods Test O (time O), Test 1 (after 6 months from production date and Test 2 (one year after the production date). During these periods the kits was stored at 15 – 30$^{o}$C. Used in this study were two control solutions, AFSC and AFSA2, and two different normal patients (containing traces of HbF, HbA1 and HbA2).

Present six months stability meets the acceptance criteria, based on the parameters tested, for the kits when stored at 15 – 30$^{o}$C for both Alkaline and Acid Hemoglobin kits.  Expiration dating will be monitored and increased as acceptance criteria allow.

*d. Detection limit:*

A commercially available abnormal control with known concentration values was serially diluted. Hemoglobin fractions were measured to access the lowest level that can be detected on the agarose gel system. Results are as follows:

Total Hemoglobin Target Value = 58.4 g/L

HbF is detected in a concentration range of: 0.54 – 17.43 g/L
HbS is detected in a concentration range of: 0.54 – 8.65 g/L

Microgel Acid Hemoglobin
HbA is detected at the lowest concentration of: 0.50 g/L
HbF is detected at the lowest concentration of: 0.60 g/L
HbS is detected at the lowest concentration of: 0.52 g/L
HbC is detected at the lowest concentration of: 0.44 g/L

*e. Analytical specificity:*

Not applicable.

*f. Assay cut-off:*

Not applicable.

2. Comparison studies:

*a. Method comparison with predicate device:*

*Microgel Alkaline Hemoglobin*

The study was performed on a total of one hundred eight (108) samples from both normal (62) and suspected pathological (46) patients. Ninety-two (92) were tested on the Interlab Microgel Hemoglobin system and with a commercially available agarose gel system, while twenty-eight (28) were compared using cellulose acetate. The samples were analyzed with the Interlab Alkaline Hemoglobin kit.
Results are as follows:

| Hb fraction | n | Slope | y-intercept | R2 |
|---|---|---|---|---|
| Hb A | 92 | 1.02 | -2.39 | 1.0 |
| Hb F | 92 | 1.05 | -1.34 | 1.0 |
| Hb S | 92 | 0.97 | 0.49 | 1.0 |
| Hb C | 92 | 1.16 | -0.31 | 0.97 |

| Hb fraction | n | Slope | y-intercept | |
|---|---|---|---|---|
| Hb A | 28 | 1.01 | -0.81 | 1.00 |
| Hb S | 28 | 1.01 | -0.13 | .99 |
| Hb C | 28 | 1.01 | 0.02 | 1.00 |

*Microgel Acid Hemoglobin*

Hemoglobin samples from thirty-one samples, either normal or with hemoglobin disorders were tested. The samples were tested with another commercially available electrophoretic system and agarose gel system. The results revealed no false negative or false positive bands by visual inspection. The study resulted in 100% agreement to the reference method for the observed bands.

The blood samples and their diagnostic assessment used for all studies were provided by hospitals outside of the United States.

*b. Matrix comparison:*

Not applicable.

3. Clinical studies:

   *a. Clinical Sensitivity:*

   Not applicable.

   *b. Clinical specificity:*

   Not applicable.

   *c. Other clinical supportive data (when a. and b. are not applicable):*

4. Clinical cut-off:

   Not applicable.

5. Expected values/Reference range:

   NOTE: values in the box refer to the Interlab Reference Ranges for Hemoglobins, analyzed by alkaline electrophoresis.

| | Interlab Reference Ranges |
|---|---|
| Hb Fraction | % |
| HbA1 | 96-99 |
| HbF | <2.0 |
| HbA2 | 1.0-3.5 |

**N. Instrument Name:**

Interlab, MICROGEL

**O. System Descriptions:**

1. Modes of Operation:

   Microgel automatically performs all the steps of the analytical procedure: application of the samples on the agarose gel plate, electrophoretic migration, gel denaruation and destaining, final gel drying.

2. Software:

   The **Elfolab/Microgel** software applications are the user interface between PC and the

Tool.

The **Elfolab** application is dedicated to the following activities:

- Display, modify, print and save the analysis result
- Start new analysis
- Insert and search patient data as well as compare with previous analysis
- Modify analysis graphs and recalculate

The **Microgel** application for the Microgel can:

- Start analysis from PC
- Show the status of the tool during the analysis
- Transfer results data to Elfolab software
- Modify the parameters and sequence of the analysis

FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

Yes ___X____ or No _____

3. Specimen Identification:

   Manual input of patient data

4. Specimen Sampling and Handling:

   Whole blood collected with EDTA is the sample of choice and should be collected following laboratory's procedure and according to the Good Laboratory Procedure Guidelines. The whole blood can be stored for one week if refrigerated at $2 – 8^{o}$ C.

5. Calibration:

   Not applicable

6. Quality Control:

   It is recommended to include in the samples the Interlab Control solutions (Hemoglobin Control AFSA2 , AFSC for Alkaline Hemoglobin testing and Hemoglobin AFSC for Acid Hemoglobin testing) in accordance with the guidelines or requirements of local, state, and/or federal regulation or accrediting organizations. In addition, the laboratory should establish acceptance parameters for each lot of control material.

**P. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:**

**Q. Proposed Labeling:**

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

**R. Conclusion:**

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.